ツリフネソウ属植物の種子の表面構造と分類

J. Phytogeogr. & Taxon.
43 : 111-117, 1995

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Systematic Implication of Seed Coat Morphology in Some *Impatiens* Species (Balsaminaceae)

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Abstract

The morphology of the seed coat surface of 21 representative species in four sections, sects. *Enantiophyllon, Microcentron, Macrocentron* and *Brachycentron* of the genus *Impatiens* sensu Warburg and Reiche was examined by scanning electron microscopy to assess the systematic relationships of the species. Similarities in seed coat surface sculpturing suggest the recognition of seven species groups, well supporting an infrageneric classification based on floral structure, seedling morphology and seed shape.

Key words : Balsaminaceae, *Impatiens,* seed coat, systematics.

The genus *Impatiens*, consisting of about 850 species, is distributed worldwide except in South America and Australia (Grey-Wilson 1980). In their earliest classification Hooker and Thomson (1859) recognized two genera in the Balsamineae, *Impatiens* and *Hydrocera*, and seven sections within the Indian *Impatiens* on the basis of habit, phyllotaxy, inflorescence and seed morphology. In 1874, Hooker proposed the division of the genus into series A (with short and ovate capsules) and B (with terete or clavate capsules). The sections used earlier (Hooker and Thomson 1859) were retained here. Thereafter, Warburg and Reiche (1895) presented a more extensive scheme for classification, recognizing two subgenera on a habit: *Acaulimpatiens* (composed only of the section *Scapigerae* Hook. f. et Thoms.), and *Caulimpatiens* (=subgenus *Impatiens*, including all the other sections of Hooker). In contrast to Hooker (1874), who did not subdivide his sections, Warburg and Reiche subdivided the subgenus *Caulimpatiens* into 12 sections on the basis of phyllotaxy, inflorescence and spur characters (Table 1). Therefore, several infrageneric taxa in Warburg and Reiche's system are partly related to different sections in

Table 1. Classification of 12 sections in *Impatiens* subgen. *Impatiens* (Warburg and Reiche 1895)

Leaves	Flowers	Spur of the lip	
		Long	Short
Opposite or whorled	Solitary or a few	*Enantiophyllon*	*Kathetophyllon*
	Many	*Choniochillon*	*Salpingochillon*
Alternate	Solitary or fascicled	*Macrocentron*	*Microcentron*
	2-5 in raceme	*Megalocentron*	*Brachycentron*
	Many in umbel	*Longicornes*	*Brevicornes*
	Many in raceme	*Longicalcaratae*	*Brevicalcaratae*

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Hooker's system. For example, section *Scapigerae* Hook. f. et Thoms. corresponds to subgenus *Acaulimpatiens* Warb.; likewise, section *Oppositifoliae* Hook. f. et Thoms. corresponds to section *Enantiophyllon* Warb., and section *Uniflorae* Hook. f. et Thoms. to section *Microcentron* Warb.

During the past few decades, studies by scanning electron microscopy have greatly contributed to systematics of various angiosperm taxa, including *Collumia* (Chuang and Heckard 1972), Cactaceae and Orchidaceae (Barthlot 1984), Bromeliaceae (Vadarajan and Gilmartin 1988), *Nama* (Chance and Bacon 1984) and *Veronica* (Juan and Fernandez 1994). As to the genus *Impatiens*, Shimizu (1979) examined the seed surface morphology of Southeast Asian species and recognized several seed types.

In this paper we report the seed coat morphology of 21 representative species in four sections of *Impatiens* sensu Warburg and Reiche and discuss its systematic value compared with the use of other morphological characters such as seed shape, number of carpels and seedling morphology.

Materials and Methods

Seeds of 21 species of *Impatiens* representing four sections, *Enantiophyllon*, *Microcentron*, *Macrocentron* and *Brachycentron* were investigated in this study. Voucher specimens are preserved in the Herbarium of Kanazawa University, Kanazawa, Japan (KANA), and the Herbarium Bogoriense, Bogor, Indonesia (BO) (Table 2). The seeds were fixed with 10% glutaraldehyde, FAA, or Telyesnicky's solution in the field and have been kept in 70% ethanol. For scanning electron microscopy, they were dehydrated through an ethanol series and dried by a critical point drier using CO_2, and then coated with gold.

Results and Discussion

Results of this investigation are summarized in Table 3. Scanning electron micrographs of the seeds of all 21 species are presented in Figs. 1-36. The seeds of the species examined are morphologically very diverse. They range approximately from 1 mm to 4 mm in length; black or brown in color; globose or flattened, elliptical or oblong in shape; and sulcate or non-sulcate.

Shimizu (1979) examined the surface sculpture morphology of the seeds and recognized two seed

Table 2. *Impatiens* species examined

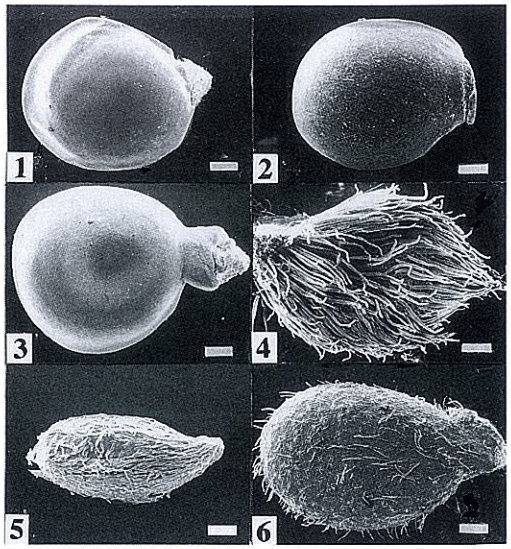
Section	Species	Locality	Voucher
Enantiophyllon	*I. chinensis* L.	Thailand: Loei	KANA 190740
	I. gardneriana Wight et Arn.	India; W. Ghats	KANA 194740
	I. griffithii Hook. f.	Malaya: Pahang	KANA 190762
	I. kleinii Wight et Arn.	India: W. Ghats	KANA 194742
	I. masoni Hook. f.	Thailand: Petchabun	KANA 189638
	I. platypetala Lindl.	Indonesia: W. java	BO 114839
Microcentron	*I. alboflava* Miq.	Indonesia: W. Sumatra	KANA 189413
	I. arriensii (Zoll.) T. Shimizu	Indonesia: E. Java	BO 20911
	I. balsamina L.	Cultivated	None
	I. bunnackii T. Shimizu	Thailand: Loei	KANA 189551
	I. charanii T. Shimizu	Thailand: Saraburi	KANA 189567
	I. hongsonensis T. Shimizu	Thailand: Maehonson	KANA 189546
	I. kerriae Craib.	Thailand: Chiangmai	KANA 189623
	I. larsenii T. Shimizu	Thailand: Surathani	KANA 189594
	I. macrosepala Hook. f.	Thailand: Phangnay	KANA 189850
	I. psittacina Hook. f.	Thailand: Surathani	KANA 189805
Macrocentron	*I. patula* Craib.	Thailand: Chiangmai	KANA 189920
	I. santisukii T. Shimizu	Thailand: Chiangmai	KANA 189742
	I. violiflora Hook. f.	Thailand: N. Tak	KANA 189917
Brachycentron	*I. noli-tangere* L.	Japan: Nagano	KANA 189559
	I. textori Miq.	Japan: Ishikawa	KANA 182684

types in section *Enantiophyllon*, five types in section *Microcentron*, and three types in section *Macrocentron*. In addition, he recognized a single type in section *Brachycentron* and section *Longicalcaratae*, respectively. However, he did not refer to correlations between the seed type and other morphological characters.

In the present study, we found that the species could be divided into seven groups on the basis of seed shape and seed coat features. These seven groups are explained below and discussed following Warburg and Reiche's system (Tables 1 and 3).

1. Section *Enantiophyllon* : Six species belonging to this section were examined and are divided into two groups :

Group I, comprising *Impatiens chinensis* (Fig. 1) , *I. kleinii* (Fig. 2) and *I. masoni* (Fig. 3) , is characterized by black globose seeds with glabrous and a shining surface, and thus comparable to the "Chinensis type" (Shimizu 1979). These three species share opposite leaves, but are dif-



Figs. 1-6. Scanning electron micrographs of seeds of *Impatiens* in section *Enantiophyllon*. 1. *I. chinensis*, 2. *I. kleinii*, 3. *I. masoni*, 4. *I. gardneriana*, 5. *I. griffithii*, 6. *I. platypetala*. Scale = 330 μm.

Table 3. Comparisons in seed and other morphological characters among 21 species examined of *Impatiens*

Section	Species	Species group	Seed shape	Surface	Seed type[1]	Carpel[2]	Seedling[3]
Enantiophyllon	*I. chinensis*	I	Globose	Glabrous and shining	Chinensis	5	F
	*I. kleinii**	I	Globose	Glabrous and shining	Chinensis	5	
	I. masoni	I	Globose	Glabrous and shining	Chinensis	5	H
	*I. gardneriana**	II	Elliptical and flattened	Pilose with long hairs	Platypetala	5	
	I. griffithii	II	Elliptical and flattened	Pilose with long hairs	Platypetala	5	E
	I. platypetala	II	Elliptical and flattened	Pilose with long hairs	Platypetala	5	E
Microcentron	*I. balsamina*	III	Globose	Verrucose	Balsamina	5	C
	*I. alboflava**	IV	Elliptical	Granulate and finely granulate	Psittacina	4	
	*I. hongsonensis**	IV	Elliptical	Granulate and finely granulate	Psittacina	4	
	I. kerriae	IV	Elliptical	Granulate and finely granulate	Psittacina	4	B
	*I. larsenii**	IV	Elliptical	Granulate and finely granulate	Psittacina	4	B
	I. psittacina	IV	Elliptical	Granulate and finely granulate	Psittacina	4	
	I. arriensii	V	Oblong elliptical and sulcate	Pilose with ventrifixed hairs	Macrosepala	4	A
	*I. bunnackii**	V	Oblong elliptical and sulcate	Pilose with ventrifixed hairs	Macrosepala	4	A
	*I. charanii**	V	Oblong elliptical and sulcate	Pilose with ventrifixed hairs	Macrosepala	4	A
	I. macrosepala	V	Oblong elliptical and sulcate	Pilose with ventrifixed hairs	Macrosepala	4	A
Macrocentron	*I. patula**	VI	Ovate and flattened	Pilose and finely granulate	Violiflora	5	
	*I. santisukii**	VI	Ovate and flattened	Pilose and finely granulate	Violiflora	5	C
	I. violiflora	VI	Ovate and flattened	Pilose and finely granulate	Violiflora	5	C
Brachycentron	*I. noli-tangere**	VII	Elliptical	Reticulate and granulate	Noli-tangere*	5	C
	*I. textori**	VII	Elliptical	Reticulate and granulate	Noli-tangere*	5	B

*Species investigated for the first time as to seed coat surface 1. See Shimizu (1979). 2. See Shimizu (1987). 3. See Shimizu (1982, 1985).

ferent in seedling morphology. *I. chinensis* belongs to "subgroup F" (cotyledon epigeal, hypocotyl not elongated and cotyledons on ground surface), while *I. masoni* is assigned to "subgroup H" (cotyledons hypogeal and swollen, Shimizu 1982, 1985).

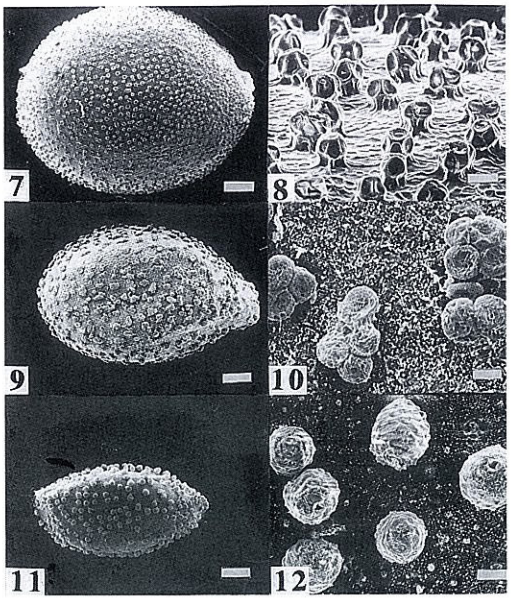
Group II, including *Impatiens gardneriana* (Fig. 4), *I. griffithii* (Fig. 5). and *I. platypetala* (Fig. 6), is characterized by ovate and flattened seeds densely covered with long hairs on the surface. But, in *I. platypetala* as shown in Fig. 6, many hairs fell off, and their scars were observed. This type was referred to the "Platypetala type" (Shimizu 1979). These three species have whorled leaves and a similar type of seedling morphology "subgroup E" (cotyledon epigeal, hypocotyl definitely elongated, epicotyl elongated; leaves of the seedlings opposite, at least by the 6 th leaf stage, Shimizu 1982, 1985).

All the species in both group Ⅰ and group Ⅱ have a perennial habit and 5-carpellate ovaries. The similarities in the seed coat surface, together with their phyllotaxy and seed shape suggest that section *Enantiophyllon* can be divided into two groups. In contrast to the similar seedling type shared by the species of group Ⅱ, the species of the group Ⅰ have two different types. Hypogeal germination of *I. masoni* is considered correlated to the tuberiferous habit, while *I. chinensis* and *I. kleinii* have a rhizomatous habit.

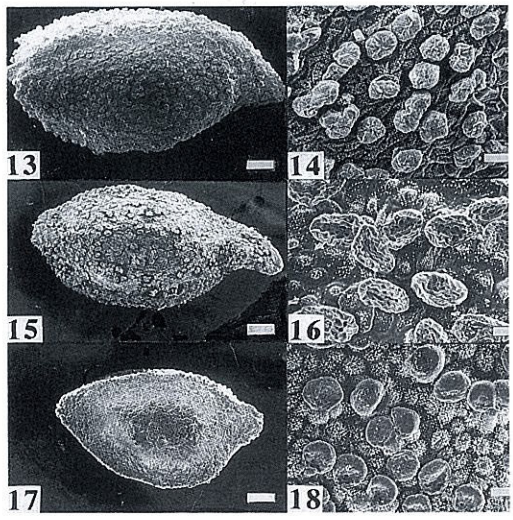
2. Section *Microcentron* : Ten species of this section were examined and are divided into three groups :

Group III. Only *Impatiens balsamina* (Figs. 7, 8) is assigned to this group. It is characterized by large globose seeds covered with small peculiar wart-like outgrowths of geometric pattern, about 50 μm in height. This type was named the "Balsamina type" (Shimizu 1979). The seedlings belong to "subgroup C" (epicotyl elongated up to nearly as long as hypocotyl; leaves of seedling all or partly opposite, Shimizu 1982, 1985). The species is also characterized by inflated tomentose capsules. Such a kind of capsule is not seen in any other groups.

Group IV, which includes *Impatiens alboflava* (Figs. 9, 10), *I. larsenii* (Figs. 11, 12), *I. hongsonensis* (Figs. 13, 14), *I. kerriae* (Figs. 15, 16) and *I. psittacina* (Figs. 17, 18), is characterized by elliptical seeds with both kinds of projec-



Figs. 7–12. Scanning electron micrographs of seeds of *Impatiens* in section *Microcentron*. 7, 8. *I. balsamina*, 9, 10. *I. alboflava*, 11, 12. *I. larsenii*. Whole seed. Scale = 330 μm.
Surface sculpturing. Scale = 33 μm.

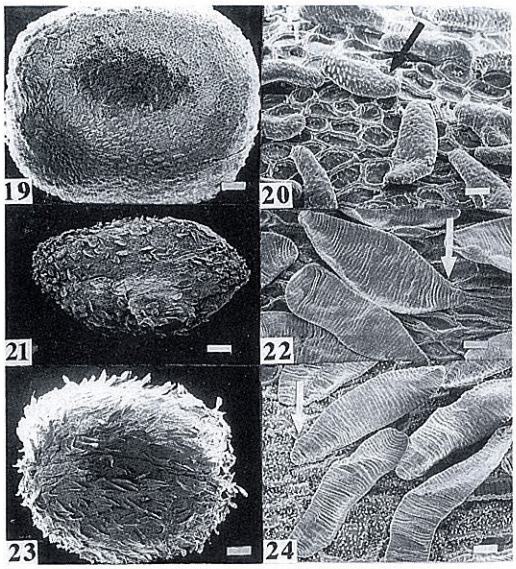


Figs. 13–18. Scanning electron micrographs of seeds of *Impatiens* in section *Microcentron*. 13, 14. *I. hongsonensis*, 15, 16, *I. kerriae*, 17, 18. *I. psittacina*. Whole seed. Scale = 330 μm. Surface sculpturing. Scale = 33 μm.

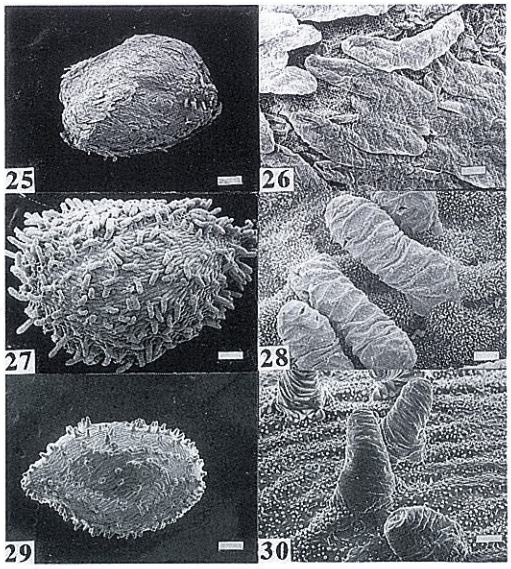
tions; big and minute. Here we use the words "granulate" and "finely granulate" for these projections (Table 3). In *I. alboflava* and *I. psittacina* 2-5 granules form a cluster, while in the other species are composed of a single form. *I. alboflava* and *I. larsenii* are quite peculiar to the genus in having non-pendulous flowers and asymmetrical wing petals (Utami and Shimizu, unpublished). However, the seed coat is covered with clustered granules in the former, while the granules on the latter are solitary. In this respect, the seed character can be used to discriminate these two species, too.

The leaves of these species are alternate, and their seedlings (except *I. psittacina*) belong to "subgroup B" (epicotyl much shorter than hypocotyl; leaves of seedling all alternate), while that of *I. psittacina* belongs to "subgroup C" (epicotyl elongated up to nearly as long as hypocotyl; leaves of seedling all or partly opposite, Shimizu 1982, 1985).

Group V, which comprises four species here, *Impatiens arriensii* (Figs. 19, 20), *I. macrosepala* (Figs. 21, 22), *I. bunnackii* (Figs. 22, 23) and *I. charanii* (Figs. 25, 26) is characterized by oblong elliptical and sulcate seeds covered with ventrifixed hairs on the surface. However, in the first two species there are no minute granules, whereas in the other two species there are numerous minute granules on the seed surfaces. In this group, therefore, the seed coat surface is diversified and it is possible to divide into "2 subgroups".



Figs. 19-24. Scanning electron micrographs of seeds of *Impatiens* in section *Microcentron*. 19, 20. *I. arriensii*, 21, 22, *I. macrosepala*, 23, 24. *I. bunnackii*. Whole seed. Scale = 330 μm. Surface sculpturing. Scale = 33 μm. Arrow shows ventrifixed point of hairs.



Figs. 25-30. Scanning electron micrographs of seeds of *Impatiens* in section *Microcentron*. 25, 26. *I. charanii*, and in section macrocentron 27, 28. *I. patula*, 29, 30. *I. santisukii*. Whole seed. Scale = 330 μm. Surface sculpturing. Scale = 33 μm.

The leaves of these four species are alternate. Their carpels are 4 in number (cf. Shimizu 1987). The seedlings belong to "subgroup A" (hypocotyl elongated, epicotyl not elongated; cotyledonary blade cornute beneath, Shimizu 1982, 1985). Ecologically the two groups (Groups Ⅳ and Ⅴ) are unique in the genus, mostly occupying sunny or shady habitats in limestone areas. The seeds of groups Ⅲ, Ⅳ and Ⅴ of section *Microcentron* are easily distinguishable by the shape of their hairs. As well as on the basis of their floral anatomy and seedling morphology, it is considered that section *Microcentron* should be separated at least into three groups.

The minute granules observed both in groups Ⅱ of section *Enantiophyllon* and Ⅳ of section *Microcentron* are considered to contain the same constituents (see group Ⅵ).

3. Section *Macrocentron* : Three species belonging to this section have been examined here. As far as these three species are concerned, their seed coat all possess the same type.

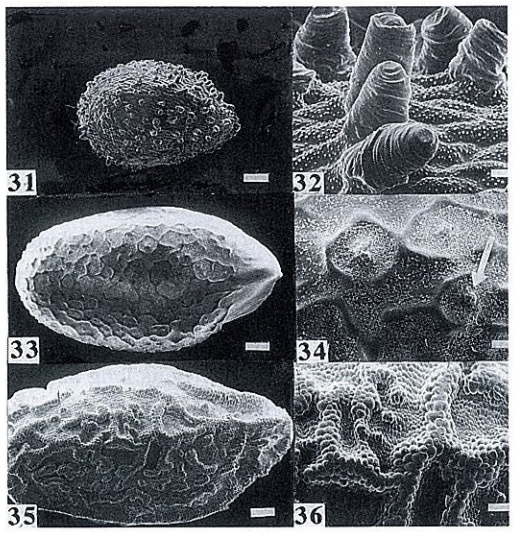
Group VI, comprising *Impatiens patula* (Figs. 27, 28), *I. santisukii* (Figs. 29, 30), and *I. violiflora* (Figs. 31, 32), is characterized by small, 1-2 mm long, ovate and flattened seeds with two kinds of projections: pilose hairs and fine granules. These seeds can be compared to the "Violiflora type" (Shimizu 1979). The minute projection of *I. violiflora* is not the production of an excretion of any kind of wax. The result of X ray microanalysis suggests that it is rich in silicon content. Its chemical composition is similar to that of helical thickenings of the large projection (Shimizu 1979). The seedlings in all the species mentioned above belong to the "subgroup C" (the first and second leaf is opposite, but the other leaves are alternate, Shimizu 1982, 1985). In this group, the seedling type well correlated to the seed coat type.

4. Secion *Brachycentron* : Two species were examined in this section and similar seed surfaces were recognized.

Group VII, including *Impatiens textori* (Figs. 33, 34) and *I. noli-tangere* (Figs. 35, 36) is characterized by elliptical seeds with reticulate surface. Epidermal cells are observable on the reticulation thickenings. *I. textori* has two kinds of granule, big and fine: 4-6 big granules (granule-like epidermal cells) formed a cluster in the center of the reticulation and a lot of fine granules are formed inside the reticulation. In *I. noli-tangere* granule-like epidermal cells form reticulation and no fine granules are found. Although these two species were put in the same group, they might be separable. Furthermore, evidence from leaf margin, flower colour and spur shape also supported that *I. noli-tangere* and I. *textori* can be divided into two distinct groups as mentioned by Hong and Oh (1993). Both of these two species have alternate leaves and similar structure of flowers, but their seedling morphology is also different. The seedlings of *I. noli-tangere* belongs to "subgroup C" (the first and second leaf of seedling are opposite, but the other leaves are alternate), and that of *I. textori* to "subgroup B" (leaves of seedling all alternate, Shimizu 1982, 1985). These facts might suggest that they are distantly related.

Conclusion

The examination of *Impatiens* seeds by scanning electron microscopy confirms the general value of seed coat features as systematic indicators. In some sections in a traditional system of the genus *Impatiens* (Warburg and Reiche 1895), seed coat features clearly partition examined species into distinctive groups as in sect. *Enantiophyllon* and sect. *Microcentron*, while they seem well to support the delimitation of other sections as exemplified by section *Macrocentron*.



Figs. 31–36. Scanning electron micrographs of seeds of *Impatiens* in section *Macrocentron*: 31, 32. I. *violiflora* and section *Brachycentron*: 33, 34. *I. textori*, 35, 36. *I. noli-tangere*.
Whole seed. Scale = 330 um Surface sculpturing. Scale = 32: 33 μm, 34: 86 μm, 36: 130 μm. Arrow indicating a cluster of granule-like epidermal cells.

Such grouping of *Impatiens* species is not always supported well by evidence from the seedling morphology, contrary to other morphological characters: phyllotaxy, floral structures, spur characters and carpel numbers. The seeds characters will give good information to establishing of the modern taxonomic system of the genus *Impatiens*.

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摘　要

ツリフネソウ属植物の属内における種間関係を分類学的に整理する目的で、ツリフネソウ属の4つの節（*Enantiophyllon, Microcentron, Macrocentron*, および*Brachycentron*）を代表する21種について、その種皮の形態の走査型電子顕微鏡による観察を行った。結果として、種皮表面の形態に基づいて7つの種群に分けることができた。この種皮表面の形態に基づく分類は、花の構造、実生形態、および種子の形にもとづく属内分類の結果とほとんどの部分で一致した。

(received May 01, 1995; accepted December 24, 1995)

○ Iwatsuki, K. *et al.* (eds.) **Flora of Japan Vol. I. Pteridophyta and Gymnospermae** A5判，302頁. 1995年11月20日，講談社. 35000円.

旧臘上記の本が届けられた。IIIaが1993年，IIIbが1994年出版だからこのシリーズは年1冊のペースで進んでいることになる。この本はシダ・裸子植物篇で，残りはIIの双子葉植物（離弁花類），IVの単子葉植物である。執筆者はシダ植物が岩槻邦男，加藤雅啓の両氏，裸子植物は山崎敬氏である。スタイルはすでにIIIで周知されているところであるが，科名（あれば、当科の主要な文献)・属名（あれば、当属の主要な文献)・種への検索表・種名・異名（ともに、出典を引用)・和名・記載（あれば、主要な化学成分)・染色体数・国内の分布・全体の分布域・主要な植物図および植物写真掲載文献の順で記述され、必要に応じて最後にノートをつけるという方式である。シダ植物にも裸子植物にも科への検索表はない。このシリーズは，小笠原諸島・琉球諸島を含めた日本全国（南千島を含む）の維管束植物のわが国初の英文版フロラとして企画，刊行されているものであり，したがって日本のフロラの研究には不可欠の文献である。この本では最新の学問的成果を取り入れつつ，シダ植物は34科94属634種216雑種，裸子植物は8科18属42種3雑種に整理されていて，両群の日本におけるフロラの規模や内容を詳細に知ることができる。難をいえば，種の記述が必ずしも検索表順でもアルファベット順でもなく全巻一貫しておらず，また，学名の著者名の引用のしかたや人名に基づく種小名の表し方が正しくなされていないなど全巻不統一な点がまま目につくことだろう。（清水建美）